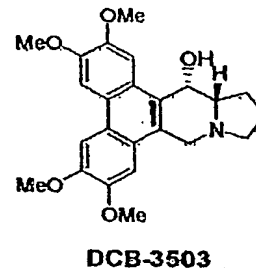
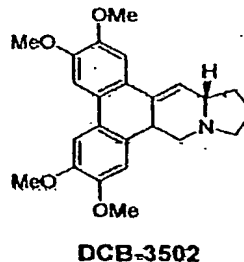
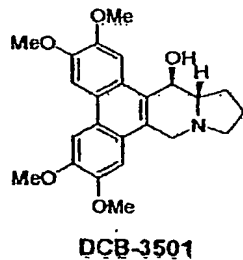
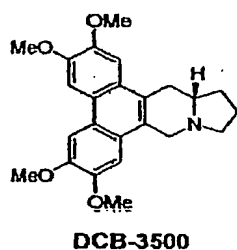


FIGURE 1

Table 1. Growth inhibition and loss of clonogenicity of KB (human nasopharyngeal carcinoma) and HepG2 (hepatocyte carcinoma) cells.

A. Chemical structure



B. EC50 (growth inhibition)

cell line	DCB-3500 (μM)	DCB-3501 (μM)	DCB-3502 (μM)	DCB-3503 (μM)
KB	0.012 ± 0.003	0.106 ± 0.084	0.234 ± 0.091	0.028 ± 0.004
HepG2	0.011 ± 0.004	0.110 ± 0.045	0.264 ± 0.115	0.035 ± 0.005

C. LD50 (colony forming ability)

cell line	DCB-3500 (μM)	DCB-3501 (μM)	DCB-3502 (μM)	DCB-3503 (μM)
KB	0.83 ± 0.31	3.07 ± 1.17	6.29 ± 3.74	1.55 ± 0.47
HepG2	0.04 ± 0.006	0.21 ± 0.13	0.45 ± 0.13	0.17 ± 0.08

FIGURE 1 CONT'D

Table 2. EC₅₀ of DCB-3500, 3501, 3502 and 3503 on the growth inhibition of KB cells and its drug resistant cells.A. EC₅₀

Cell line	DCB-3500 (μM)	DCB-3501 (μM)	DCB-3502 (μM)	DCB-3503 (μM)
KB	0.012±0.003	0.106±0.084	0.234±0.091	0.028±0.004
KB-MDR	0.014±0.005	0.143±0.082	0.282±0.138	0.026±0.008
KB-7D	0.012±0.007	0.225±0.206	0.527±0.080	0.045±0.010
KB-7D-Rev	0.011±0.006	0.070±0.057	0.289±0.188	0.025±0.010
KB-Hu-R	0.025±0.007	0.070±0.042	0.218±0.167	0.036±0.006
KB-Hu-Rev	0.016±0.004	0.045±0.004	0.127±0.095	0.028±0.016
KB-100	0.020±0.010	0.103±0.033	0.179±0.063	0.038±0.001
KB-100-Rev	0.010±0.003	0.118±0.031	0.247±0.122	0.041±0.009

B. Description of resistant cell lines

Cell line	Biochemical changes	Resistant to
KB-MDR	gp 170 ↑	VP-16, Taxol, Adrimycin, Vincristine
KB-7D	Topo II ↓, MRP ↑	VP-16, Vincristine, Adrimycin
KB-7D-Rev	Topo II ↓	VP-16, Adrimycin
KB-Hu-R	Ribonucleotide reductase ↑ CdR kinase ↓	Hydroxyurea, AraC, Gemcitabine
KB-Hu-Rev	CdR kinase ↓	AraC, Gemcitabine
KB-100	Topo I ↓ XRCC 1 ↑	camptothecin, topotecan, SN-38
KB-100-Rev	Topo I ↓ XRCC 1 ↑	camptothecin, topotecan, SN-38

FIGURE 1 CONT'D

Table 3. Impact of DCB-3500 and 3503 on cell cycle progression

KB		G0-G1	S	G2-M
Control	24 h	75	10	15
Nocodazole		0	10	90
3500 0.03 uM		62	19	19
3500 0.1 uM		56	26	18
3500 0.3 uM		54	30	16
3500 1 uM		52	36	12
3503 0.1 uM		68	18	14
3503 0.3 uM		52	33	15
3503 1 uM		63	24	13
3503 3 uM		57	30	13

HepG2		G0-G1	S	G2-M
Control	24 h	45	27	28
Nocodazole		2	18	80
00 0.03 uM		47	29	24
00 0.1 uM		44	33	23
00 0.3 uM		51	21	28
00 1 uM		41	27	32
03 0.1 uM		50	26	24
03 0.3 uM		55	20	25
03 1 uM		53	21	26
03 3 uM		47	25	28

FIGURE 1 A

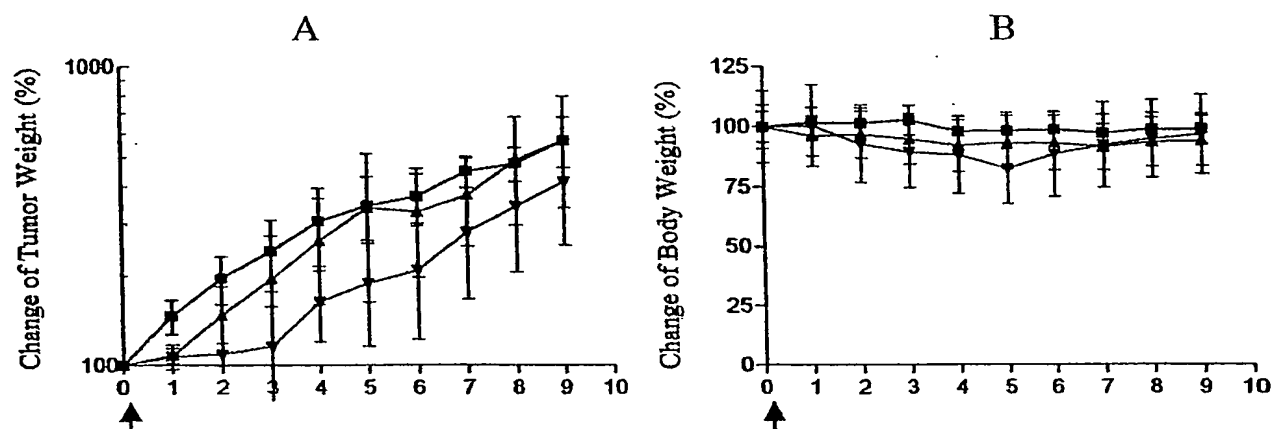


FIGURE 2

Regulation of p53 in response to conventional chemotherapeutic drugs and 3500, 3503 in KB cells

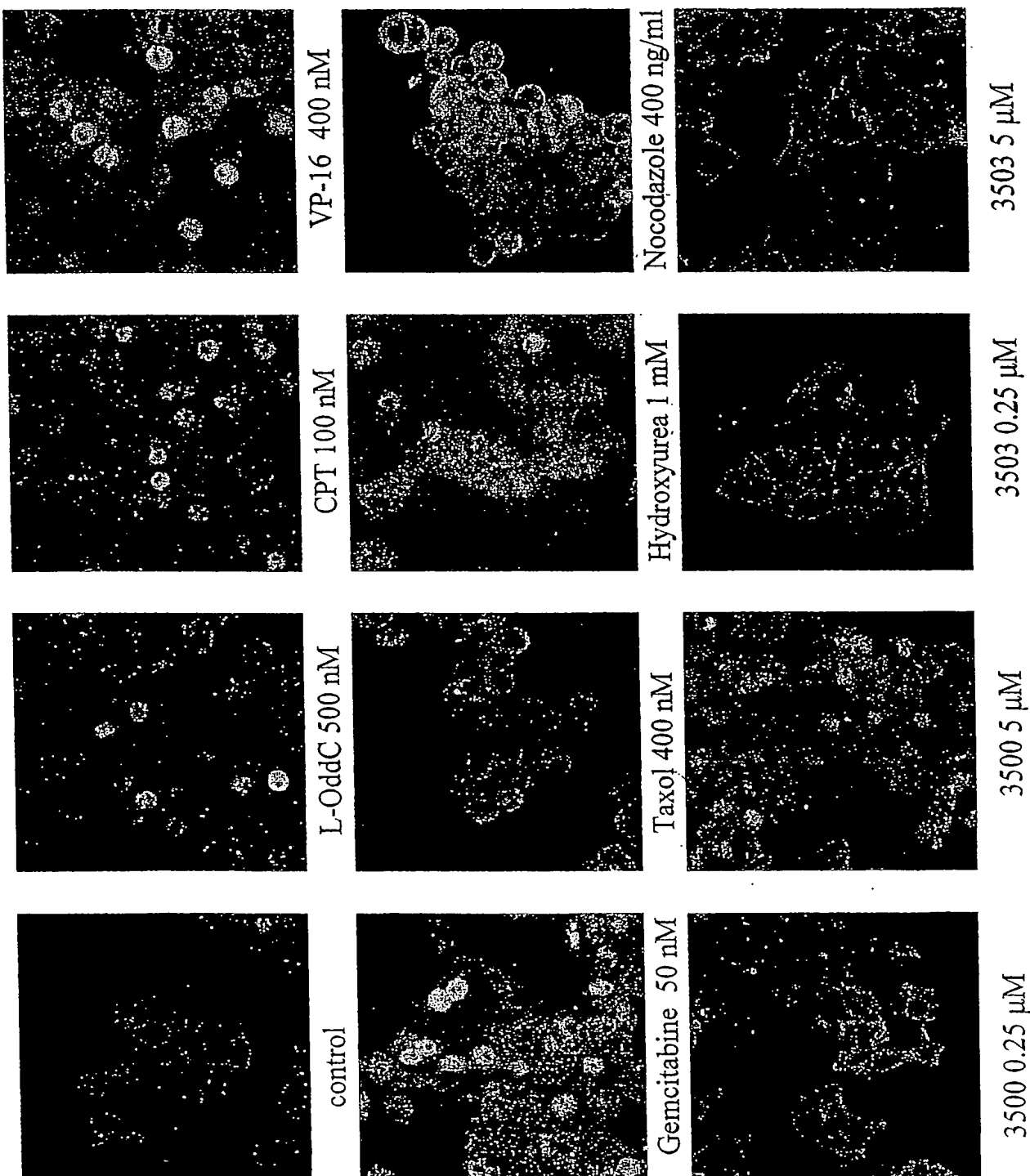
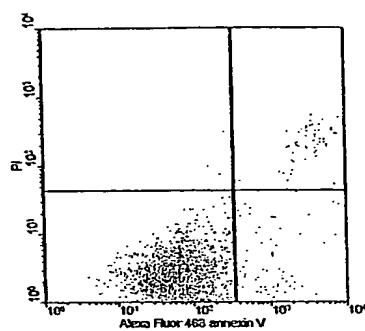
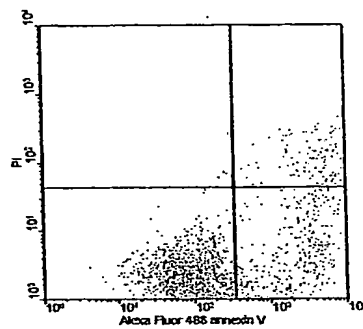
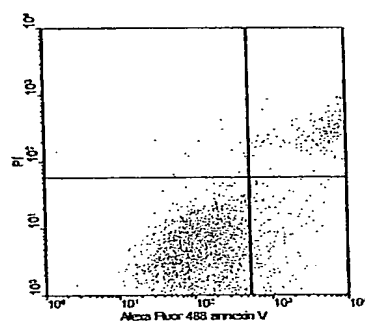


FIGURE 3



KB untreated

KB 3503 3 μ M

HepG2 untreated

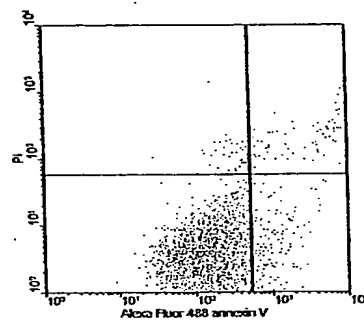
HepG2 3503 3 μ M

FIGURE 4

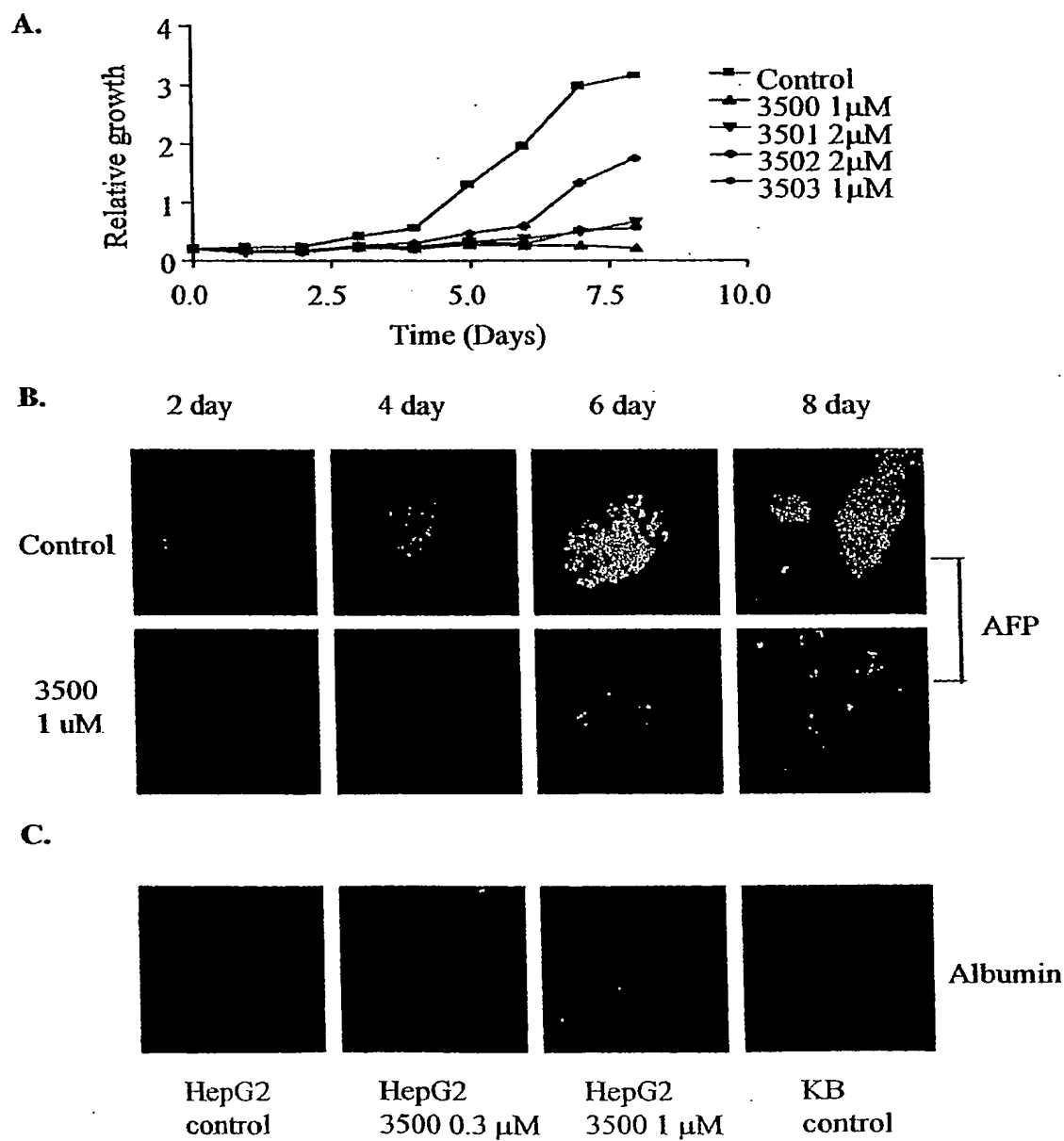
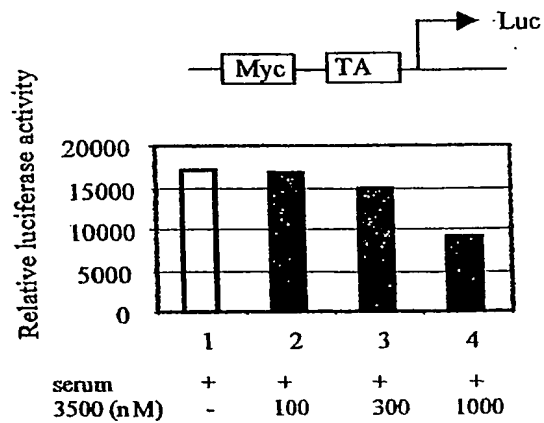
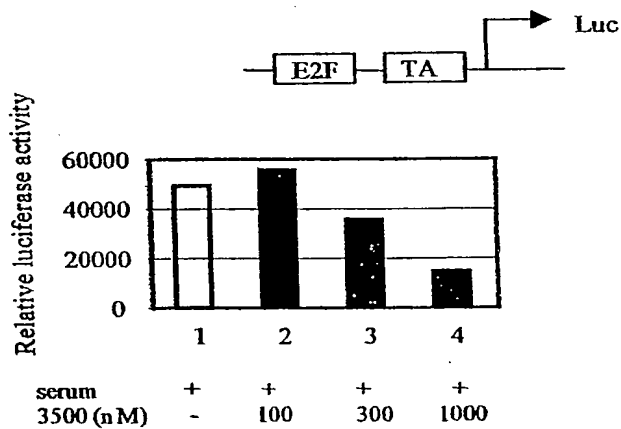


FIGURE 5

A.

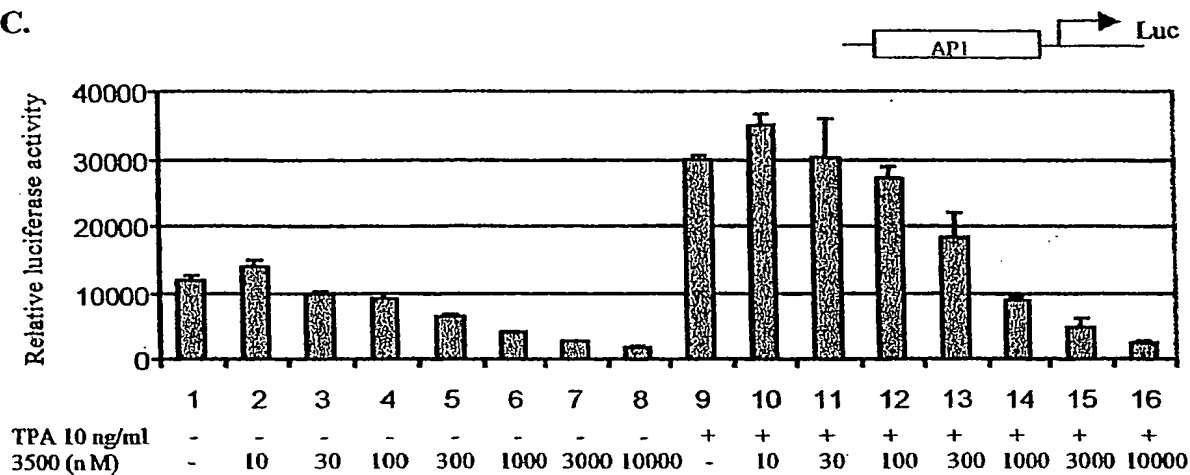


B.

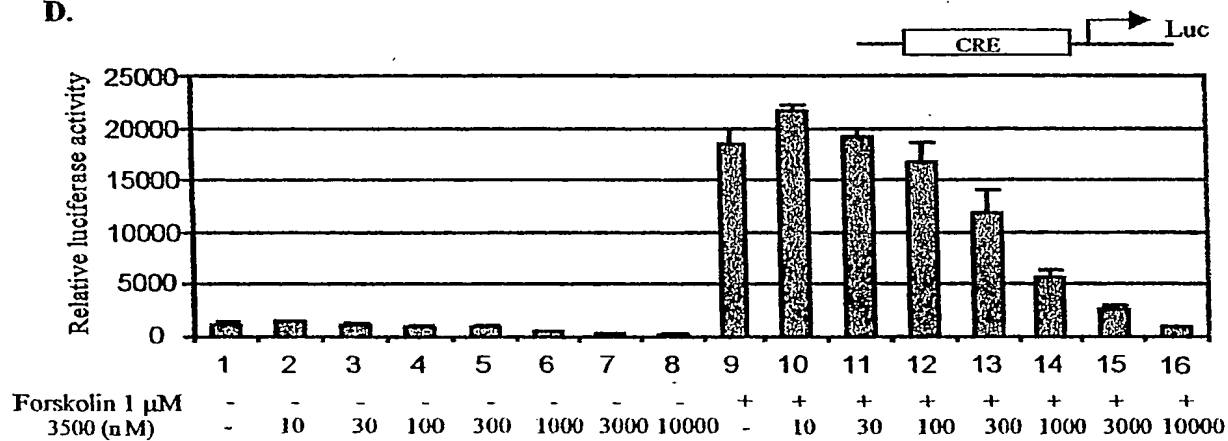


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FIGURE 5 CONT'D

C.



D.



E.

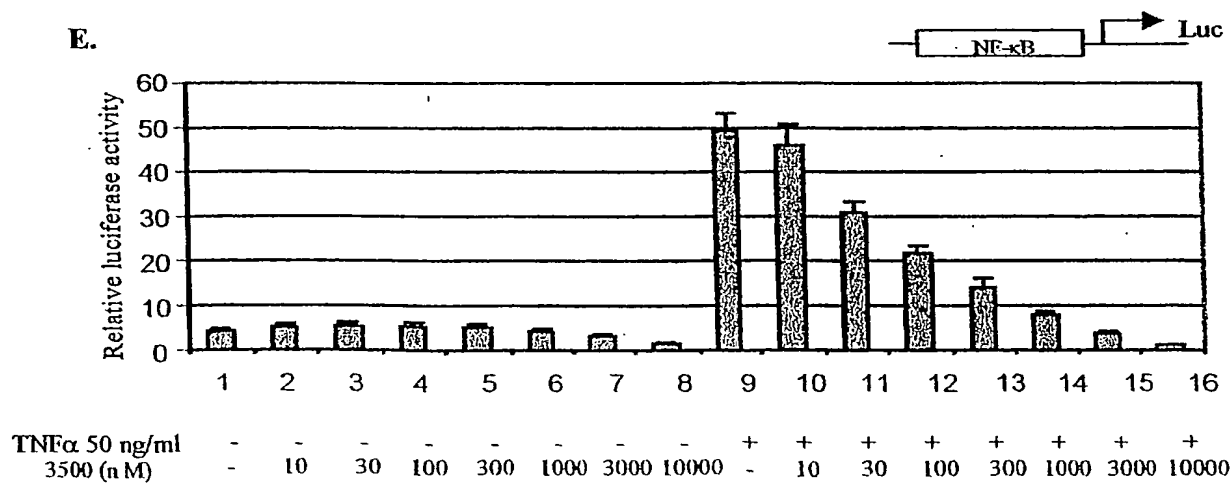


FIGURE 5 CONT'D

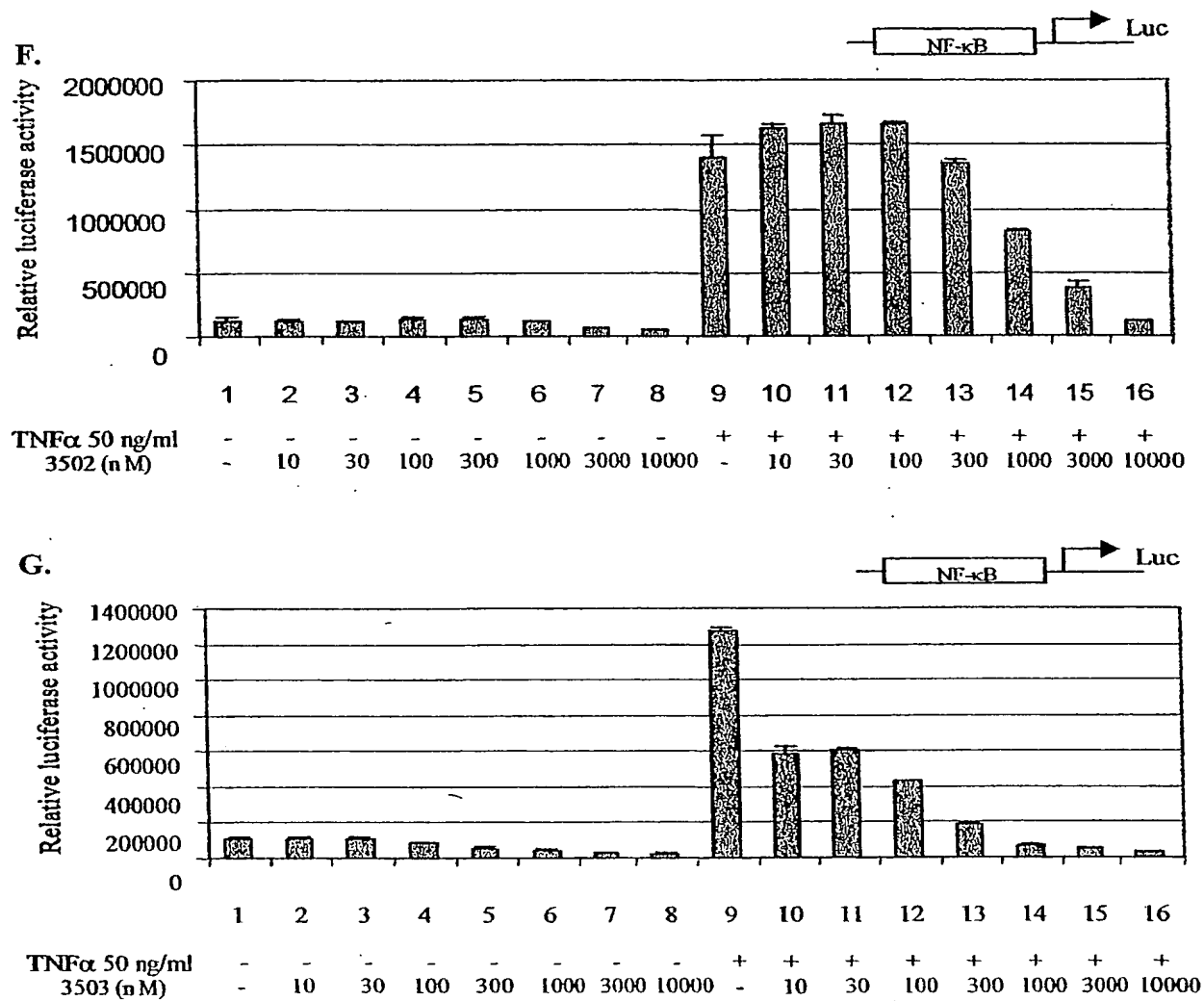


FIGURE 6

Scheme I

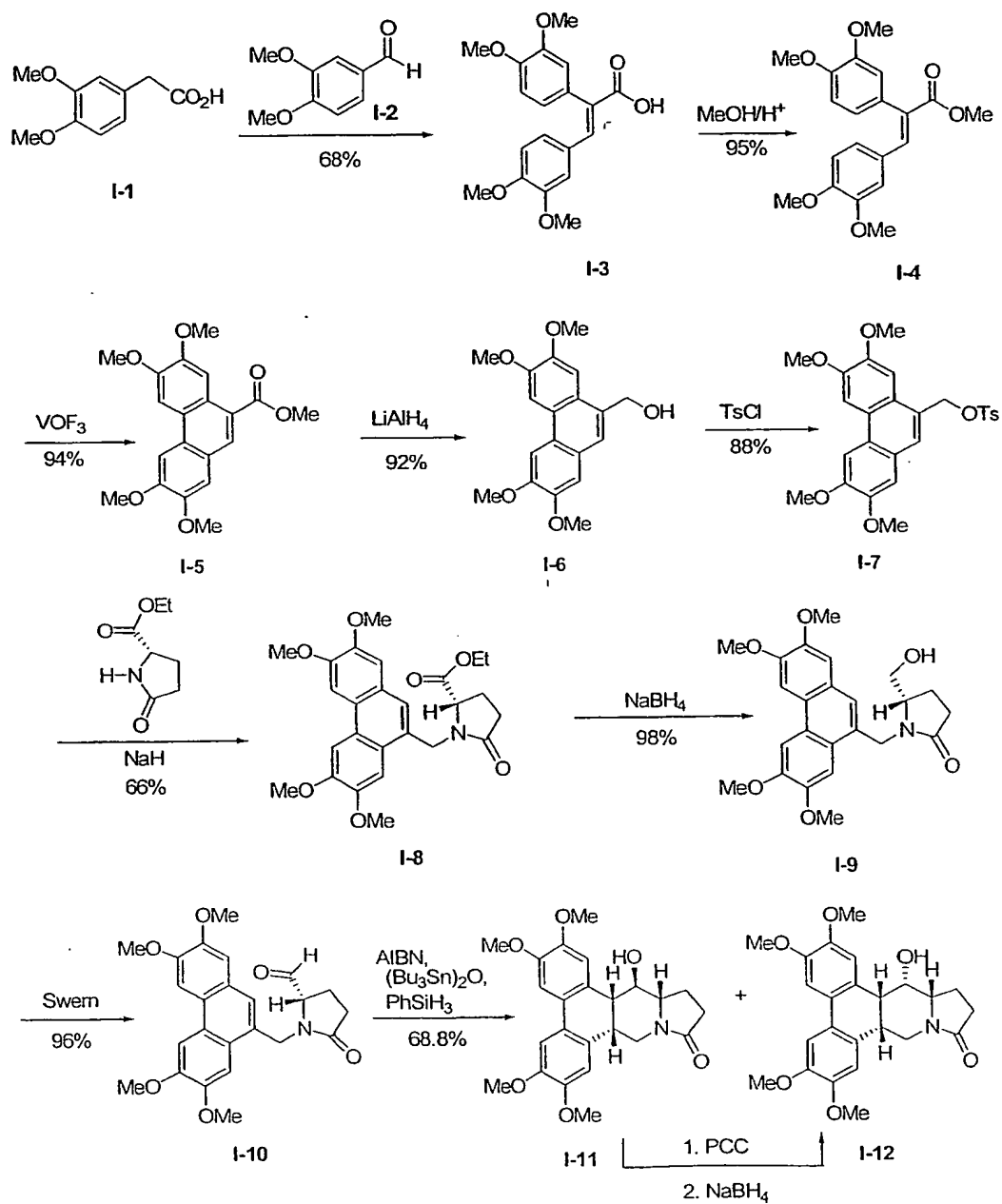
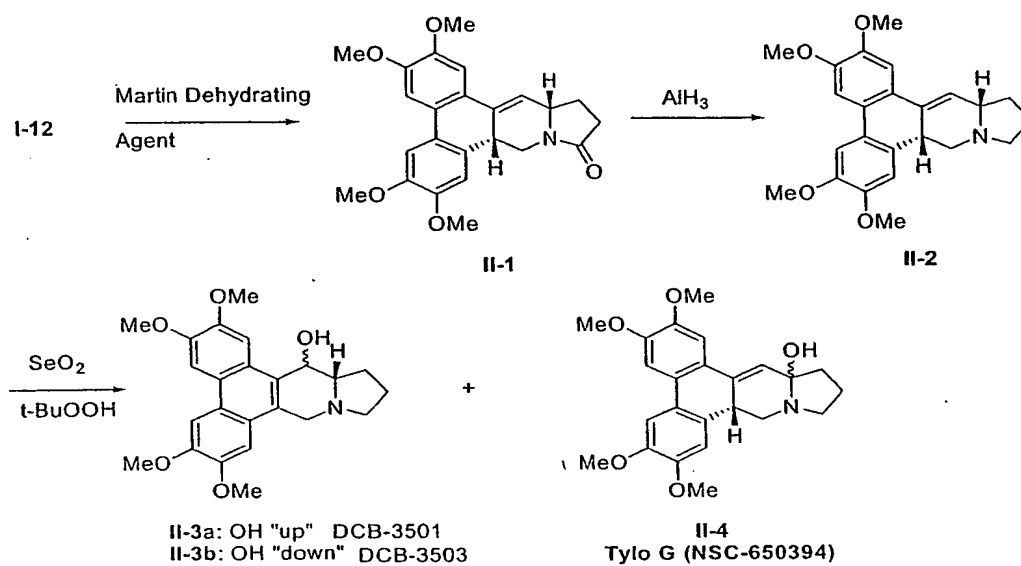


FIGURE 7

Scheme II



Scheme III

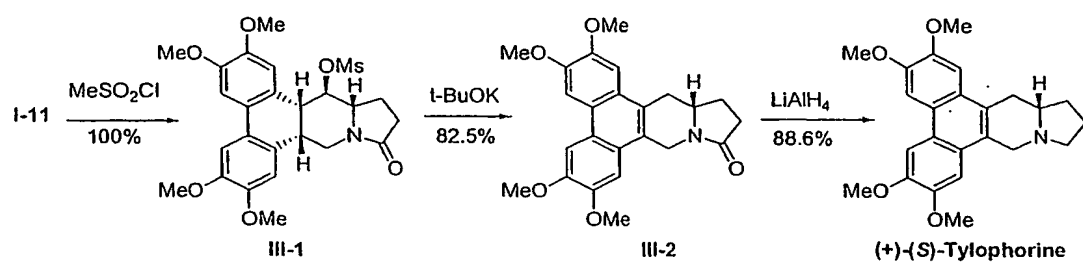


FIGURE 9

Scheme IV

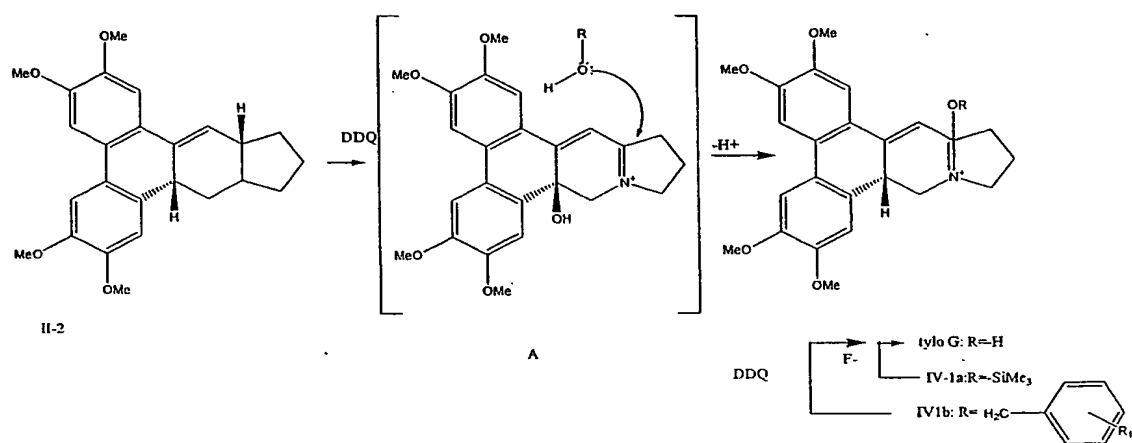


FIGURE 10

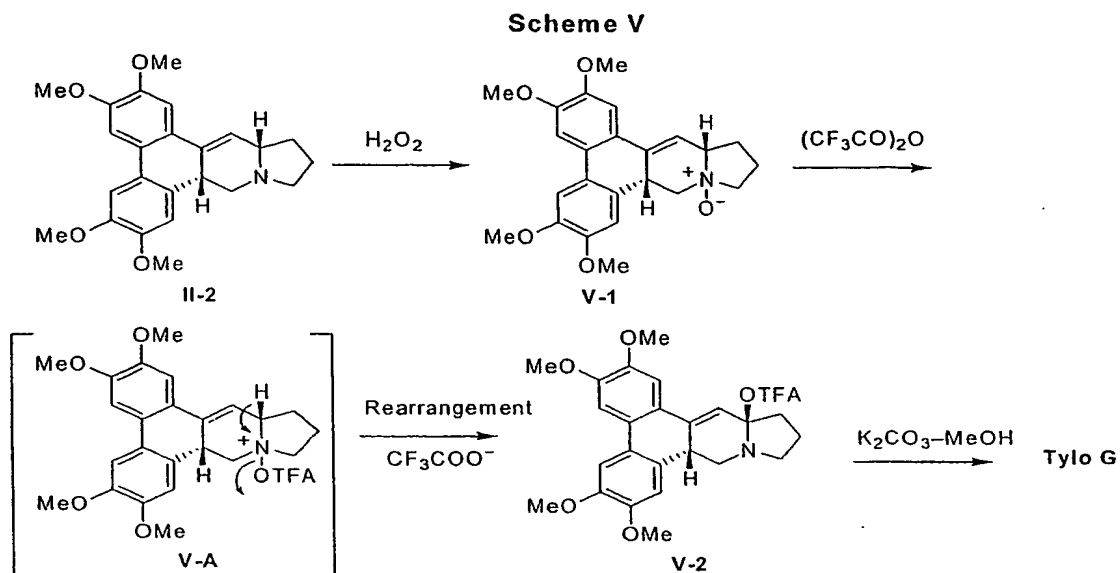


FIGURE 11

Scheme VI

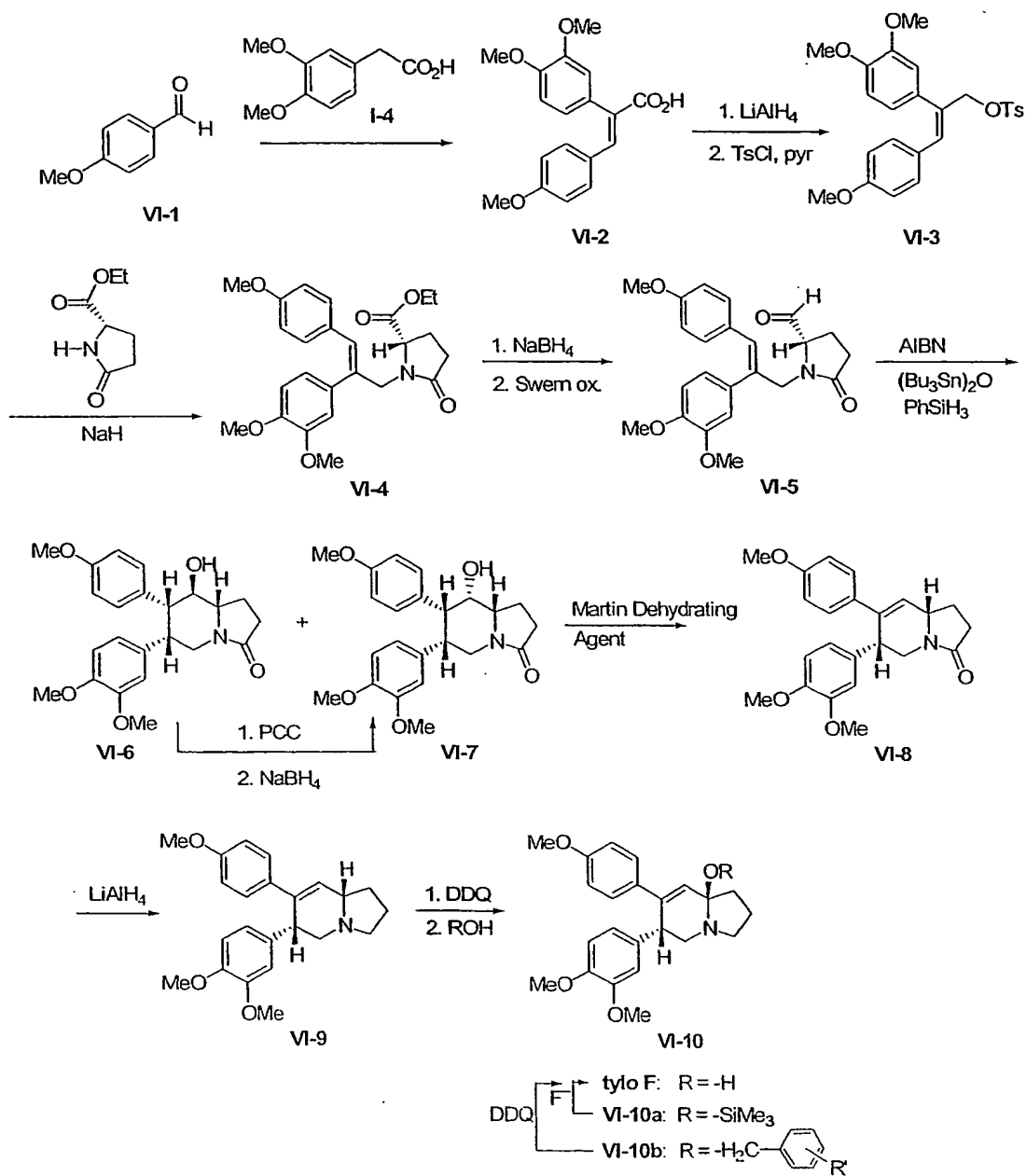


FIGURE 12

Scheme VII

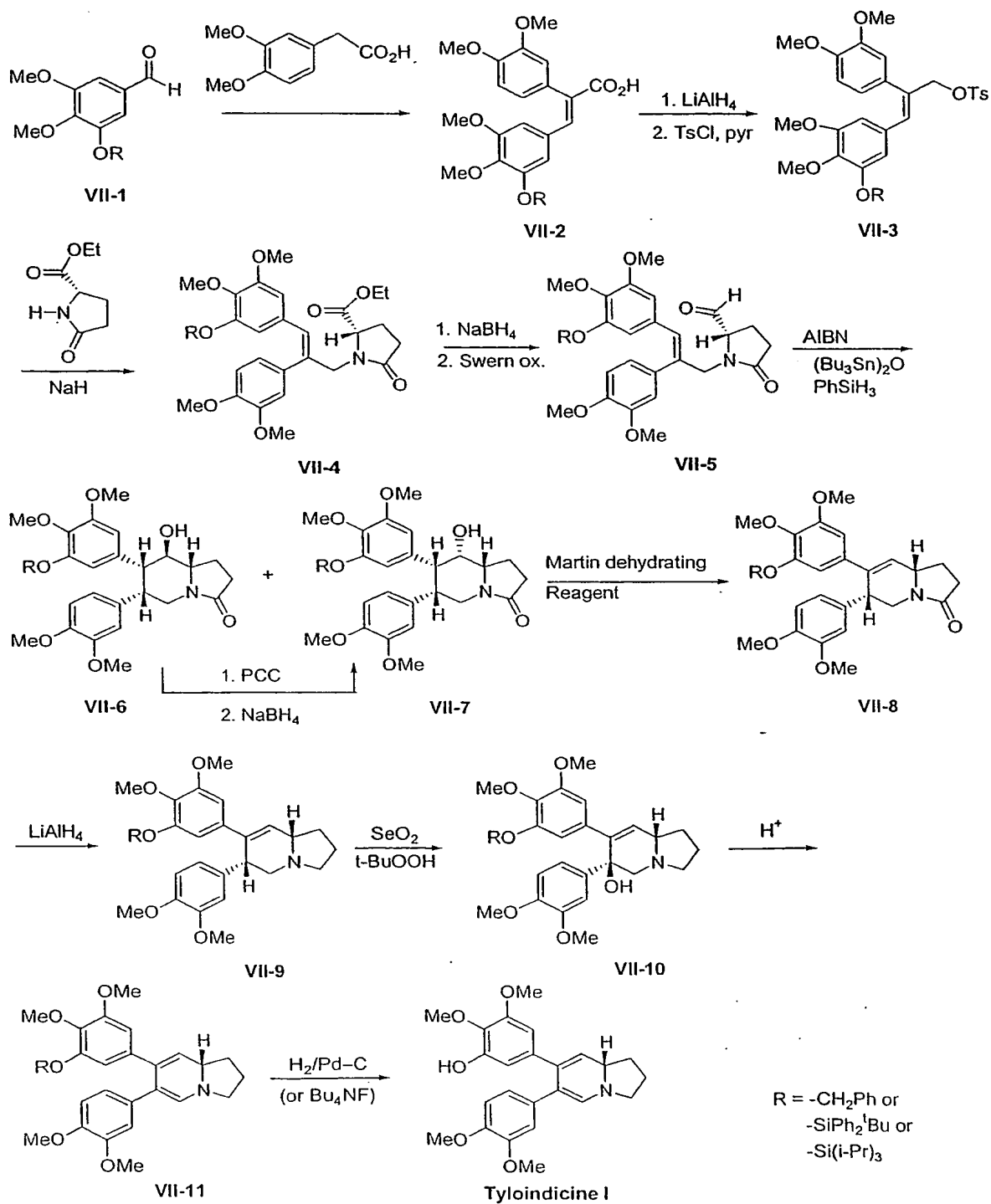


FIGURE 13

Scheme VIII

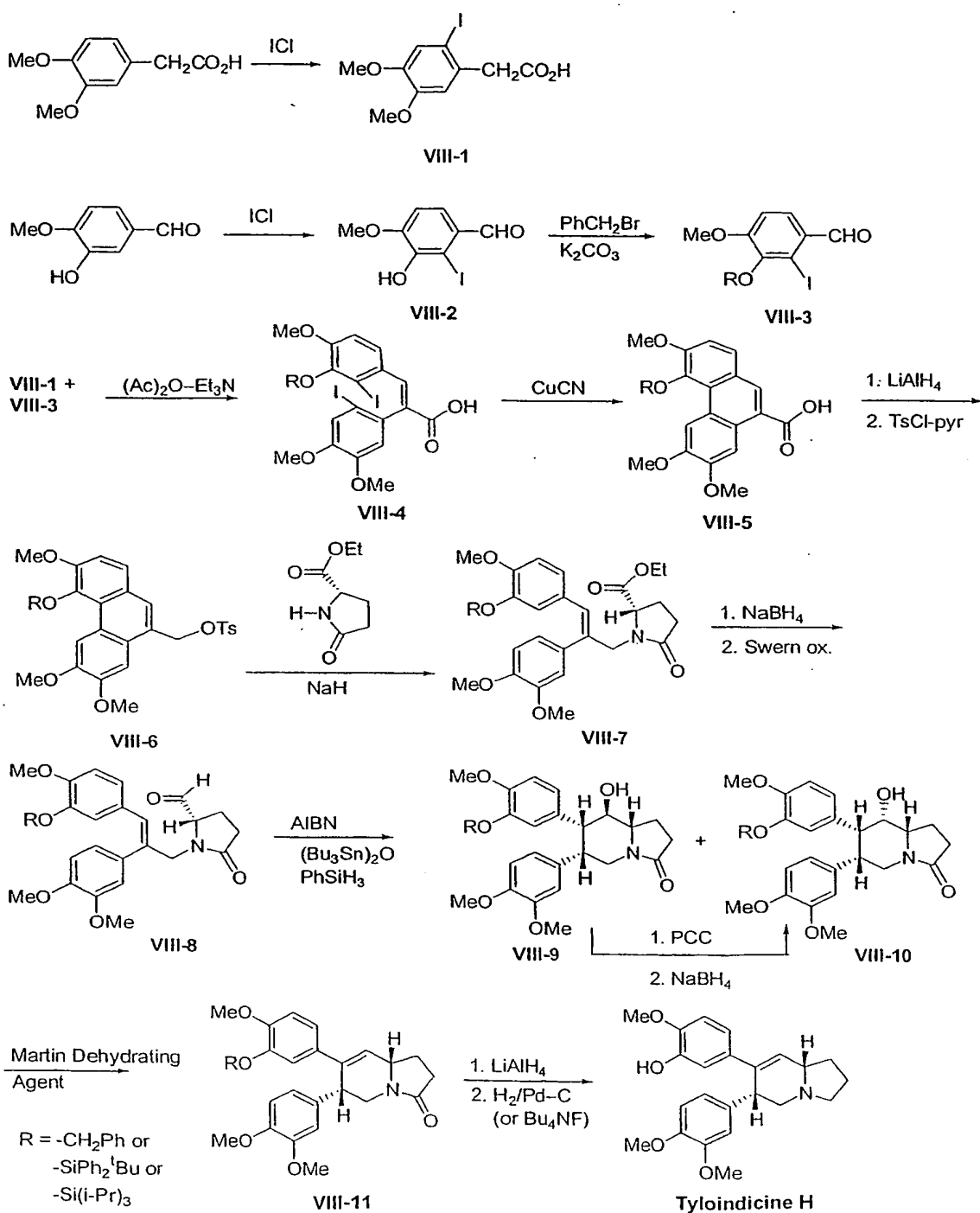
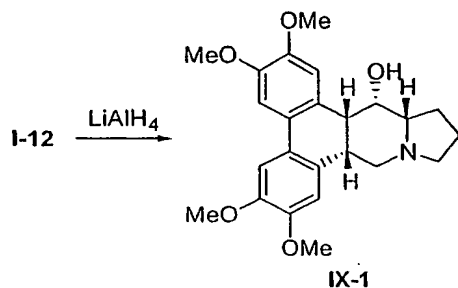
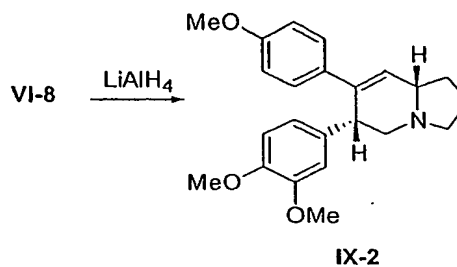


FIGURE 14

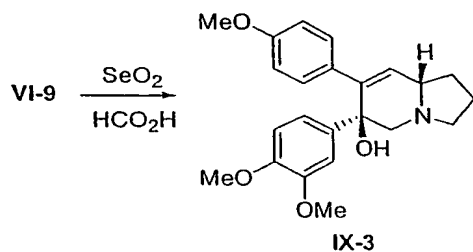
Scheme IX



Gives analogue (epimer) to NSC-717334.
Similar conversions possible for VI-6, VI-7, VII-6,
VII-7, VIII-9, VIII-10. VII-6, VII-7, VIII-9, VIII-10 can
be deprotected, then reduced for additional
analogues. A few will be made to search for activity.

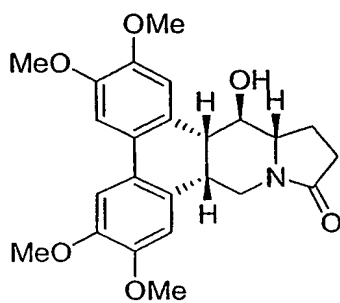


Screen VII-9 and deprotect to give analogue.



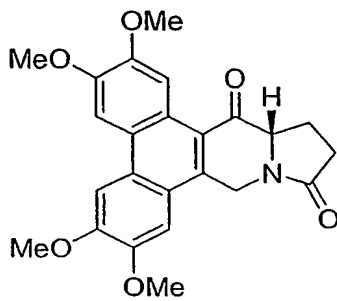
Gives the tylo F analogue of II-3 (NSC-716802,
active). VII-10, deprotect. Also LiAlH₄, SeO₂
on VIII-11, deprotect.

FIGURE 15



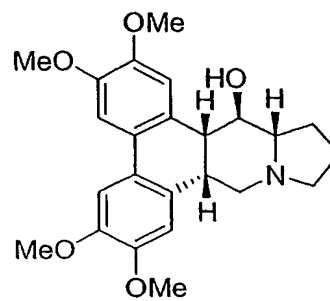
NSC-717334, I-11

(III)



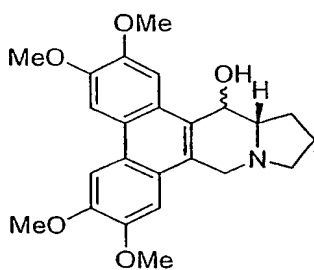
NSC-712822

(IV)



NSC-717336

(V)



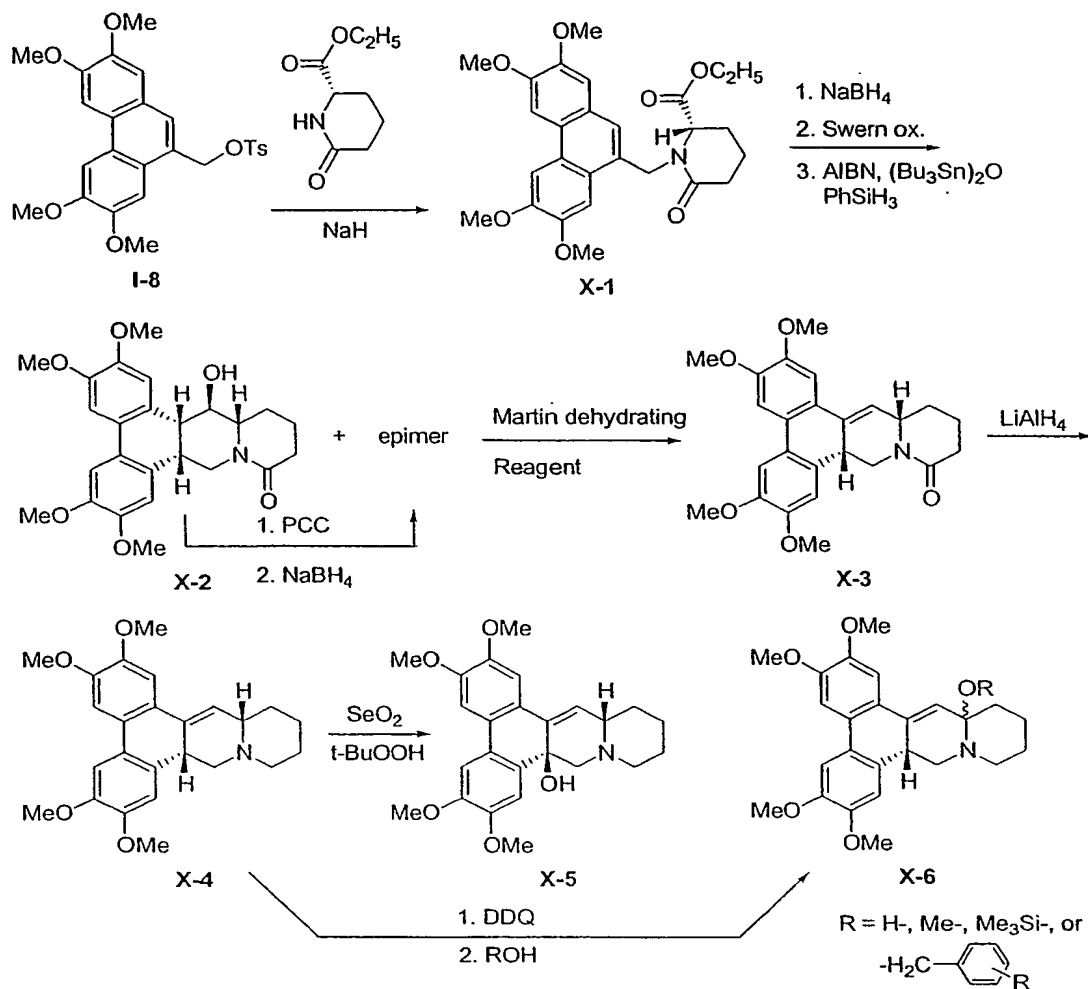
II-3a: OH "up" DCB-3501

II-3b: OH "down" DCB-3503, NSC-716802

(VI)

FIGURE 16

Scheme X



Scheme XI

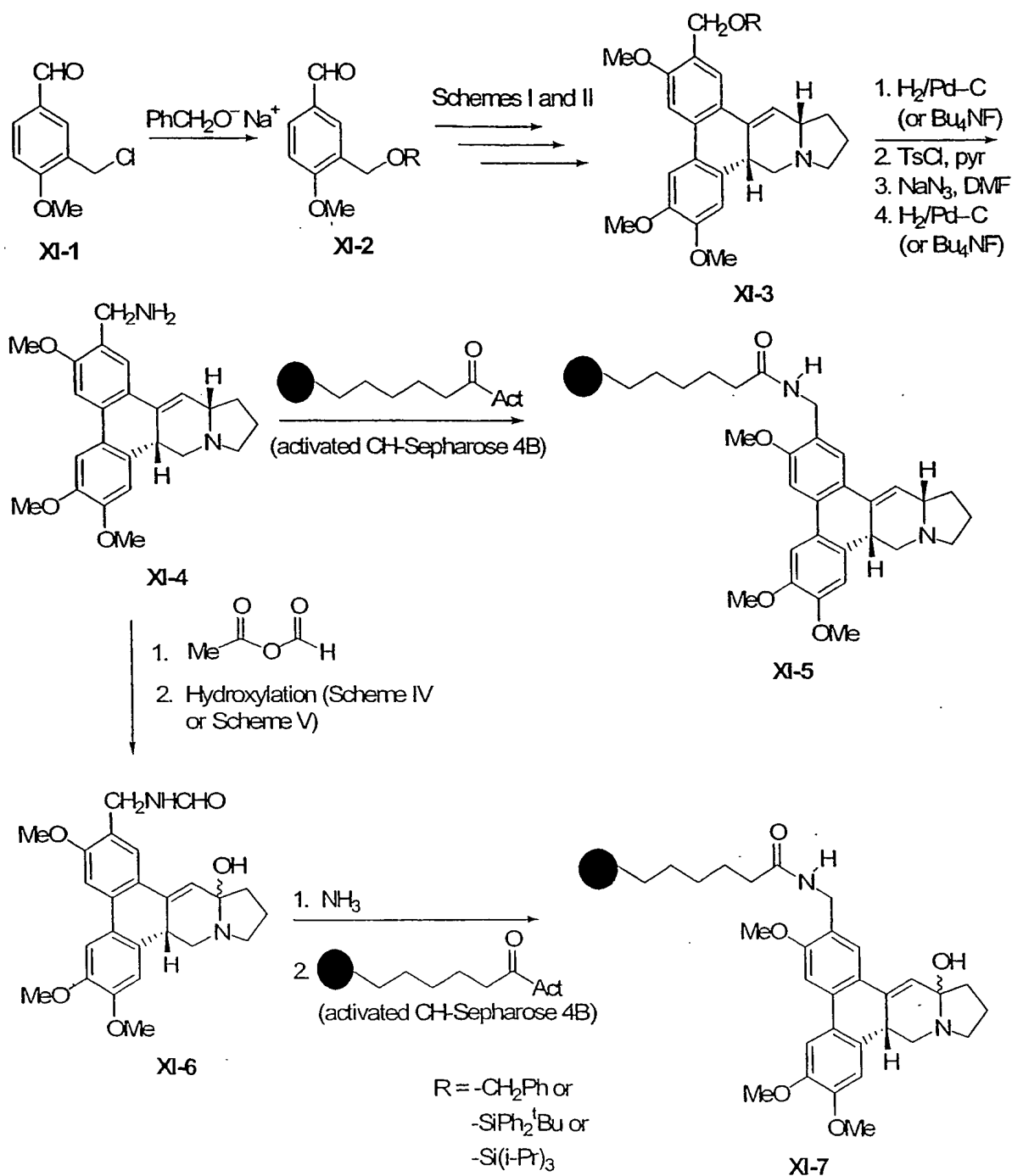


FIGURE 18

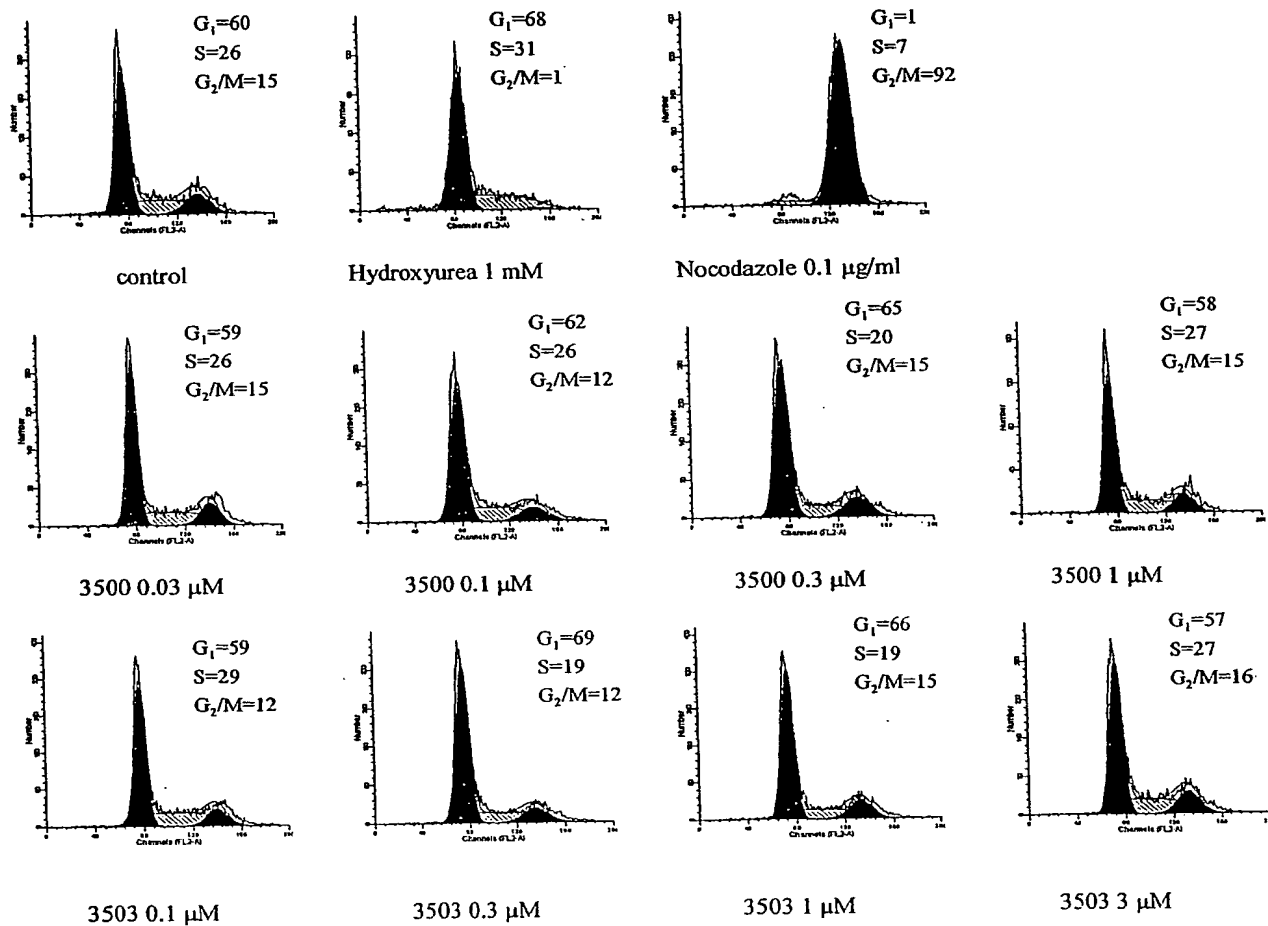
B. HepG2

FIGURE 19

Clonogenic Assays Effect of ZH-152

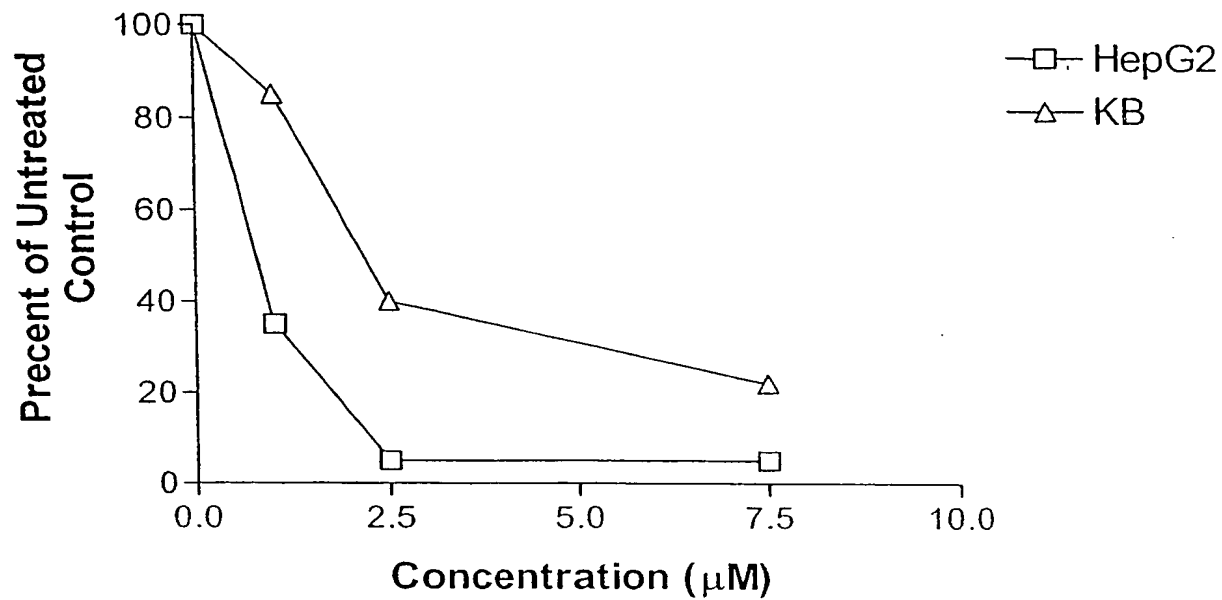
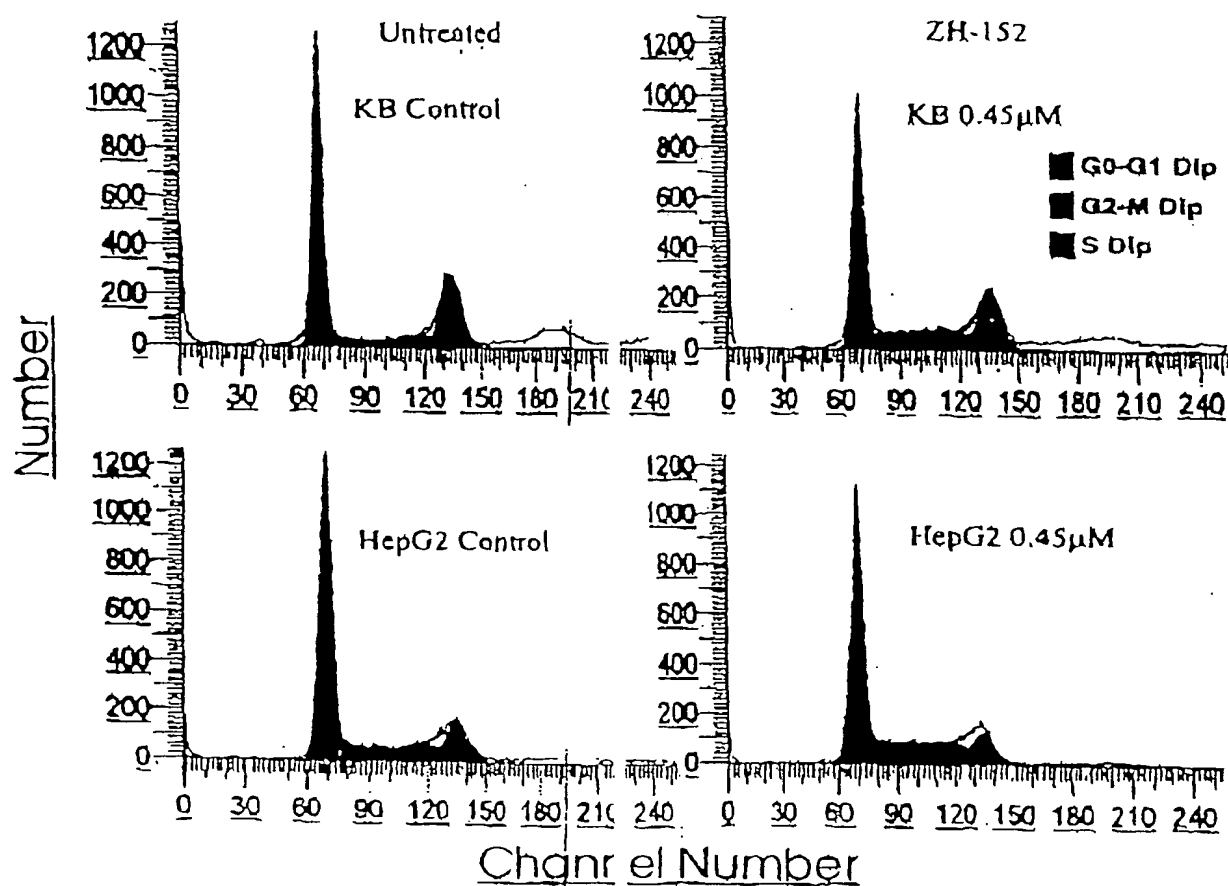


FIGURE 20

Effect of 24h Treatment with ZH-152 on Cell Cycle Progression



ZH-152 slows down the cell progress in S-phase of both cell lines. Thus, the growth inhibition of these two cell lines by ZH-152 is due to the inhibition at targets responsible for S-phase progression. Additional biochemical determinants may play a role in the preferential killing (loss of clonogenicity) of HepG2 to that of KB.

FIGURE 21

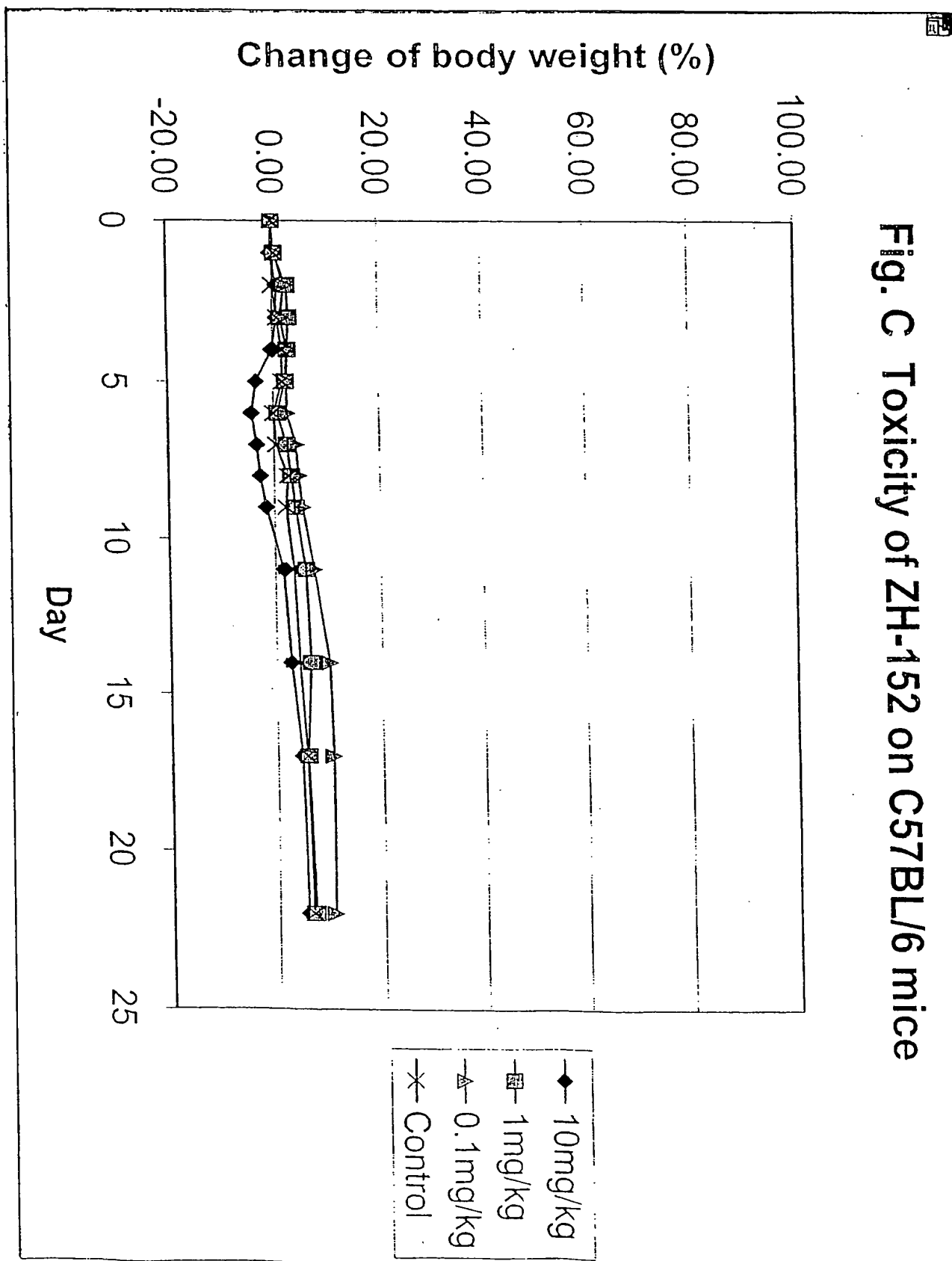


FIGURE 22

